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SEVERAL PROBLEMS OF THE METHODOLOGY OF SANITARY-MICRO BIOLOGICAL INVESTIGATION OF THE AIR IN LIVING QUARTERS

[This is a translation of an article written by A. I. Shafir in Gigiyena i Sanitariya (Hygiene and Sanitation), No 11, 1959, pages 7-11.]

No hygienic description of the air in living quarters is complete without data from microbiological analyses. However, the latter do not have the same independent importance as the results of physical or chemical analyses of the air considered in isolation.

The majority of present-day Soviet and foreign authors hold the view that sanitary-microbiological analysis of the air in living quarters should be made in the following sequence: a) determining the microbe population (the total number of microorganisms); and b) determining the concentration of hygienically significant microorganisms.

Indexes of the microbe population of the air are of themselves a fairly good indication of the extent of pollution, especially when the aspiration and sedimentation methods are used in parallel. Simultaneous determination of the index for hygienically significant microbes (also by the aspiration and sedimentation methods) considerably facilitates the hygienic and epidemiological evaluation of the atmospheric medium.

The problem as to which microorganisms are hygienically significant for the air in living quarters has not yet been fully solved. Apparently, the most correct method in making sanitary-microbiological analyses of the air in living quarters is to combine a count of the anhemolytic streptococci and streptococci viridans, ascertaining the streptococcus index. Both of these microorganisms reach the air through the human nasopharynx, thus being associated by virtue of their common origin, just as the intestine of man and that of warm-blooded animals serves as a natural reservoir for bacteria of the Bacillus coli group. Being excreted into the air with fine droplets of saliva and mucus, the anhemolytic Streptococci and Streptococci viridans are absorbed by particles of dust in the room, are easily attracted by convection air currents, are sometimes spread over considerable distances. Despite the fact that Streptococci viridans are found in the oral cavity of human beings much more often than anhemolytic Streptococci, the latter are frequently found in the dust collected

from furniture and floors, and floating in the air. This is due to the fact that anhemolytic Streptococci are more resistant to the external environment than Streptococci viridans.

The discovery of anhemolytic Staphylococci in the air is of undoubted interest; but, unfortunately, only a very few investigations have been devoted to a study of the conditions of the dissemination of this microbe in the external environment.

Sanitary-microbiological analyses made with the sedimentation (dish) method should be carried out at the same points in the room as analyses using the aspiration method. The results of analyses of the air by the sedimentation method are frequently contradictory. Placing the dishes in glass jars (so-called "vegetation vessel") 30 centimeters in height and having a diameter of 16 centimeters, makes for a certain orderliness in carrying out analyses by the sedimentation method. In the absence of such jars, Petri dishes can be placed on stands surrounded by sheathing (cylinders) made of tin, plywood, or cardboard. In using the glass jars it is also necessary to have available a simple apparatus consisting of a metal or wooden ring (on which the Petri dish is set) with a thick wire attached to it and extending beyond the edge of the dish. The individual parts of the above apparatus for making air cultures are shown in Figure 1.

The securing of somewhat more stable and mutually comparable results from analyses when the sedimentation method is rationalized (placing the dishes in jars) is due to the fact of eliminating heavy accumulations of microbes on the dish from adjacent air layers and creating the conditions for the deposition of microbes predominantly from that column of air directly above the surface of the culture medium. Employment of the method of making air cultures in dishes placed on the bottom of glass jars or on cylinders is especially indicated for rooms used by many people (theatres, schools, railroad stations, etc.).

It is advisable to give the results of air analyses by the sedimentation method in terms of the number of microorganisms deposited per square meter of surface per minute. This index is not customarily used, but it deserves consideration for the following reasons: a) the surfaces of Petri dishes vary, as we know; b) various authors have proposed various periods of time for exposing dishes in taking air cultures -- from five to 20 minutes, and even more (up to one hour); c) if the proposed units are employed (square meter per minute) the results of separate analyses can be reduced to a single yardstick, which is important for purposes of comparison.

For clean air in living quarters in the summer, the development of up to 75 colonies of microorganisms per square meter of area per minute is typical, and up to 200 colonies is typical for the winter. For impure air, there are more than 300 microorganisms per square meter per minute in the summer, and more than 500 in the winter.

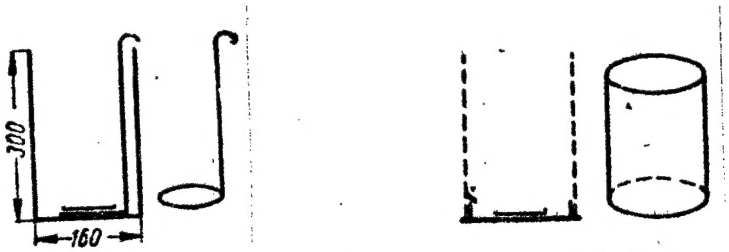


Fig. 1
Rationalization of the Sedimentation Method

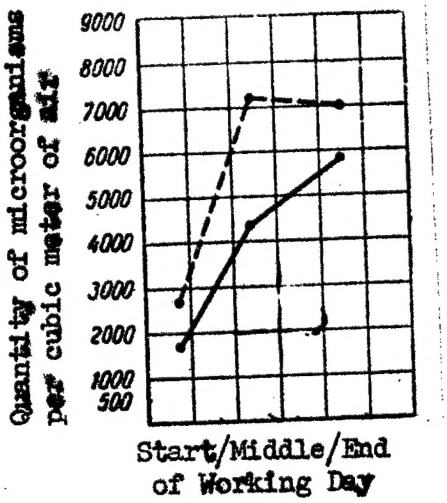


Fig. 2.
Results of Bacteriological Analysis of Air in Isolated Operating rooms (data from T. A. Krotovaya)

With respect to the exposure time for the dishes, it should be noted that the longer the exposure, the more reliable the results. If the air is relatively clean, the dishes (according to observations which have already been made) can be left exposed for as long as 20 minutes, since no difficulties are encountered in making the culture or subsequent counts of developed colonies. The average length of exposure should be ten minutes.

The aspiration methods of sanitary-microbiological analysis in use at present are relative, since they do not make it possible to trap all of the microorganisms present in any given volume of air being used for purposes of a culture. Those methods which ensure

stability of data and logically explicable patterns in the breakdown of microorganisms (e.g., by seasons of the year, or by various periods of the day) are to be preferred.

Figure 2 gives comparative data for microbiological analysis of the air in a surgical operating room for blood donors. These data were obtained by two aspiration methods -- the slot and the centrifugal. (There are several types of slotted apparatuses: the Burdillon, Lidwell and Thomas (1941), Rask and Sip (1949), Krotov (1953), Decker and Willson (1954), and others. Centrifugal apparatuses have been developed by Wells (1933) and Shafir (1939).) These data were provided by T. A. Krotova (Leningrad).

In operating rooms, the microbic impurities in the air increase constantly from the start of the working day to the end. Although the data furnished by the centrifugal method are smaller than those furnished by the slot method, they give a clearer picture of the dynamics in the quantitative breakdown of the atmospheric microflora. The aerocentrifuge makes it possible to take cultures from large volumes of air (form 0.5 to one cubic meter). It is this fact which governs the relative clarity of the results of the analyses, since atmospheric microflore are distributed through rooms very unevenly.

The vital functions of the majority of microorganisms in the air are usually depressed as a result of the effect of the light and absence of moisture in the environment. Therefore, a sensitive method is necessary in making cultures, and in the subsequent cultivation of atmospheric microbes. Unfortunately, those aspiration methods which involve drawing the air through dry and moist filters, depositing microbes on the drying surface of the culture media under the impact of an air current, etc., do not satisfy this principle.

Examination of the rather numerous investigations dealing with an evaluation of various methods of sanitary-microbiological analyses of the air reveals a considerable number of methodological errors. The most egregious of these is the practice of considering the best method to be that which makes it possible to obtain the largest numerical analytic index (which, moreover, often represents not so much the result of direct discovery of microorganisms found in the air, as a total of corresponding arithmetic calculations). Especially large (and, at the same time, very ragged) figures for bacterial polution are obtained with the so-called D'yakonov method, and in using the Rechmenskiy filters. This is due to the fact that a considerable part of the microorganisms are removed with the filtered air and the filter itself, from the volume of air being used for culture purposes, and also to the fact that a large number of multiples are introduced in calculating the results of the analysis.

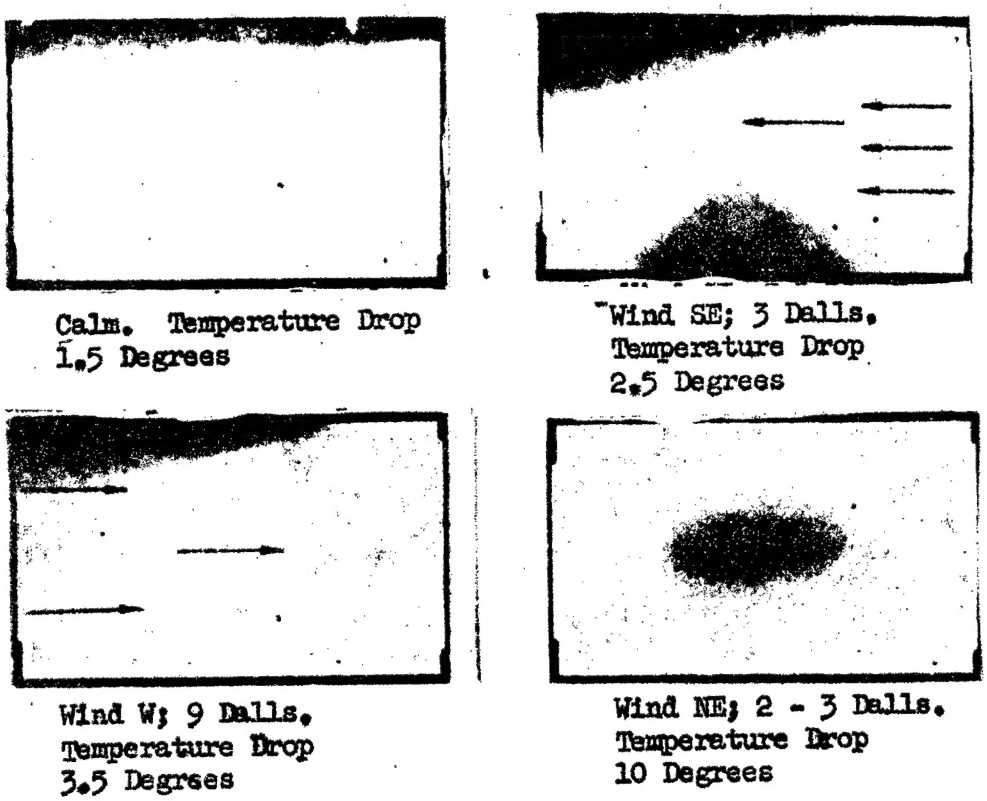
Another substantial shortcoming in these investigations is the incorrect selection of points for making cultures from the air,

without taking into account its most important property -- its dynamism -- which is due to the convection currents which are constantly being formed in buildings. The formation of air currents in a building is affected by many factors: the difference between the temperature of the air outside and that in the room; the velocity and direction of the wind; the heating system; the arrangement of heating apparatuses, windows, and doors. As is shown in Figure 3, the layer of waste air in rooms (i.e., air which is somewhat hotter and which contains relatively more moisture, carbon dioxide, dust, and microorganisms) may, depending upon the weather conditions, be distributed in widely differing parts of rooms.

The data in Figure 3 confirm the necessity for a preliminary study of the convection air currents in rooms in selecting points for making cultures from the air in the most rational manner. There are several simple and feasible methods of analyzing convection currents: a) on the basis of the deflection of very fine cigarette paper or split silk thread, fastened to a wooded or metal rod; and b) observation of the movement of artifically created fumes (dans) of ammonium chloride, stannic chloride, or other chemical compounds. (Descriptions of the fume method, and of apparatuses for producing fumes, may be found in many manuals of hygiene, heating, and ventilation.)

Points for making cultures from room air should be located where the air currents are the most evident, and where there are no currents. If it is not possible to analyze the convection currents, air cultures should be taken envelopewise; i.e., at five points in the room, located on various planes and at distance of 1.5 meters from the walls and floor. An exception should be made for the point in the middle of the room, where the culture is taken at the level of human breathing. In the event of a shortage of equipment, the analysis may be limited to three cultures taken at the following points and in the following order: a) in the middle of the room, at a height of 1.5 to 2 meters from the floor; b) just under the ceiling, near a wall on which sunlight is falling; c) on the floor, in the opposite corner of the room. It is preferable to report results for each point individually, without using average figures, since averages only complicate the picture of the distribution of impurifications polluting the air in the room.

Other mistakes in sanitary-microbiological analyses of the air include the following: a) failure to adhere to the principle of systematization in the analyses made; b) the still-extant practice of using coefficients for converting data from sedimentation analyses to volumetric measurement, despite the different colloidal nature of the microorganisms deposited from the air and the bacertial suspension phase in them; c) an incorrect attitude toward the data from the analyses; in particular, isolating average percentages from the total number of estimations, which only tend to camouflage the



Calm. Temperature Drop 1.5 Degrees

Wind SE; 3 Dalls. Temperature Drop 2.5 Degrees

Wind W; 9 Dalls. Temperature Drop 3.5 Degrees

Wind NE; 2 - 3 Dalls. Temperature Drop 10 Degrees

Fig. 3.
Position of Layers of Waste Air in a Room in Accordance with Weather Conditions

order and actual characture of the magnitudes obtained in the tests; d) working with data from analyses, without indicating in what season of the year or what hour of the day, the air cultures were taken.

The content of microorganisms in the air of a room is subject to constant variation. Different populations in the air of test rooms (depending upon the time of year or day) obviously require the application of different conditions and procedures for comparative analyses, first of all in cultures taken from large or small volumes of air. (A considerable part of the above shortcomings are exemplified by the article by V. V. Vlodavets (Gigiyena in Sanitariya, 1957, No 1). This article does not contain a single absolute figure which would make it possible to show the actual volume of the population of the air in the test room.)

The latest data indicate that clothing and bedding constitute major sources of pollution and infection of the air in living quarters. Therefore, sanitary-microbiological analyses of clothing, bedding, and other "soft" furnishings should be carried

out simultaneously with analyses of the air. Analyses of clothing and bedding follow the same system as those of the air.

The discovery of viruses in the air is assuming a very promising character. Analyses of this kind should also acquire special importance in solving problems of the prophylaxis of hospital infections, evaluating methods of disinfection and sterilization of the air, establishing the effectiveness of ventilation, etc. Aspiration apparatuses, making it possible to accumulate virus agents in liquid cultures, and a bacteriophage as a model of a filtrable virus (A. A. Gerasimenkok, Sverdlovsk) may be used for sanitary-virological analyses of the air. In recent years A. I. Shafir and A. A. Sinitskiy successfully tested paper filters of ventilating systems for retaining viruses. Also, a start has been made at the natural observation of the behavior of viruses in the air of medical institutions having wards for flu patients and others suffering with virus diseases.

Some of the most important problems now facing Soviet sanitary bacteriologists include the unification, standardization, and maximum improvement of the technique of bacteriological and virological analysis of the air, the development of basically new apparatuses, and encouraging the creativity of reationalizors and inventors.

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